AMYLOID NEUROPATHY

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ABSTRACT

Amyloidosis is the name given to a group of diseases characterized by the extracellular deposition of insoluble amyloid fibrils in different tissues and organs like kidneys, heart, liver, skin, nerves etc. Clinical manifestations of amyloidoses are determined by the amyloid precursor protein type, by the tissue containing amyloid deposits, and by the quantity of stored amyloid. We present here the data of a patient with sensory polyneuropathy, orthostatic hypotension, nephritic syndrome and benign IgG monoclonal gammapathy, in which we diagnosed amyloid neuropathy.

Key words: amiloid neuropathy, polyneuropathy, monoclonal gammapathy, histology

Amyloidosis is the name given to a group of diseases characterized by the extracellular deposition of insoluble amyloid fibrils in different tissues and organs like kidneys, heart, liver, skin, nerves etc. The importace of amyloid deposits varies in different types of amyloidoses.

Three categories of systemic amyloidoses are known:

A) hereditary diseases with autosomal dominant inheritance:

- TTR amyloidosis due to transthyretin gene mutations (chromosome 18).
- ApoAI amyloidosis due to ApoAI gene mutations (chromosome 11).
- AGel amyloidosis due to gelsolin gene mutations.

B) sporadic monoclonal immunoglobulin light chains primary amyloidosis (AL);

C) aquired amyloidosis (AA) secondary to long lasting chronical infections.

Clinical manifestations of amyloidoses are determined by the amyloid precursor protein type, by the tissue containing amyloid deposits, and by the quantity of stored amyloid.

As Benson and Kincaid (2007) have showed, reviewing the known data, the neuropathy is a major feature of several types of systemic amyloidoses, especially of TTR amyloidosis, but also of ApoAI and AGel amyloidoses (20% of the patients with AGel amyloidosis have peripheral neuropathy). Peripheral neuropathy was not reported in aquired (AA) amyloidosis.

We present the data of one of our patients (R. N.), male, aged 53, farmer, reffered to us for a sural nerve and gastrocnemius muscle biopsies by an internal medicine service. His clinical diagnosis comprised sensory polyneuropathy with orthostatic hypotension, nephritic syndrome, benign IgG monoclonal gammopathy (moderate increase of his seric IgG), and ankilopoetic spondilitis diagnosed 10 years before his presentation in our hospital. His family history declared by our patient was nonrelevant.

Six years before his presentation in our laboratory, the patient observed the insidious onset and progressive course of digestive dysfunctions, polakiuria, distal dysesthesia in his inferior and superior limbs. At presentation neurological examination

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showed distal trophic lesions in his legs, moderate predominantly distal amyotrophy in his inferior and superior limbs, decreased deep tendon reflexes and the bilateral absence of the ankle ones. A rectal biopsy showed chronical non-specific inflammatory signs.

The sural nerve biopsy showed on cryostat sections the very severe decrease of the myelinic fibre number without relationship with their size, frequent myelinic bullae, and deposits of an unstructured material pink with HE staining (figs.1 and 2), pale blue with Gomori trichrome, yellow-pink with vanGieson, and red with Congo red (fig.3) with pale birefringence in polarized light. These deposits were present under the perinerve, in and around the vessels walls, and in the endonerve dissociating the nerve fibres. On semithin sections from nerve fragments embedded in synthetic resins showed, also, (figs.4 and 5) a very severe loss of myelinic fibres – 891 fb/mm2 (N=7,000-11,000fb/mm2) with a bimodal histogram (fig.6) suggesting an interstitial neuropathy. The practically absence of amyelinic fibres, the proliferation of endoneurial collagen, as well as the presence of deposits of unstructured material could be observed too.

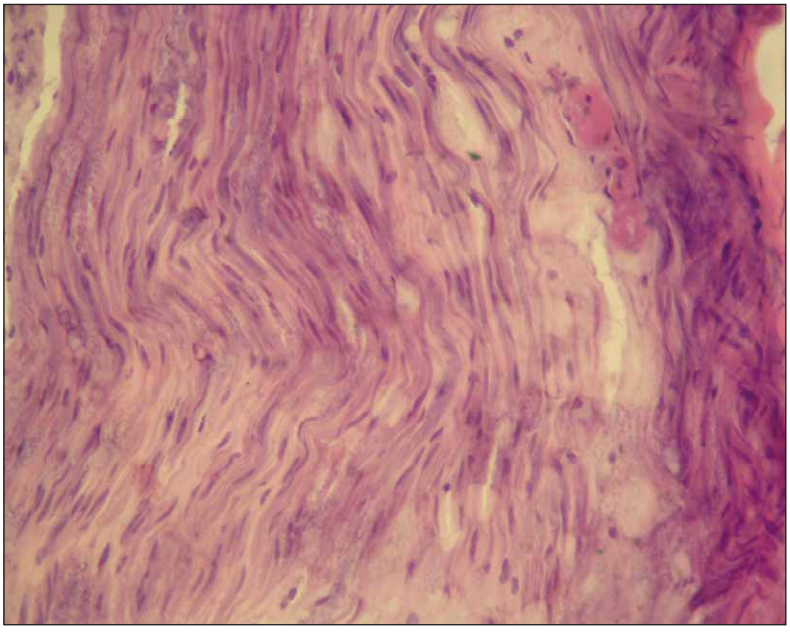

FIGURE 1. *Unstructured material under the perineurium. HE, ob.40.*

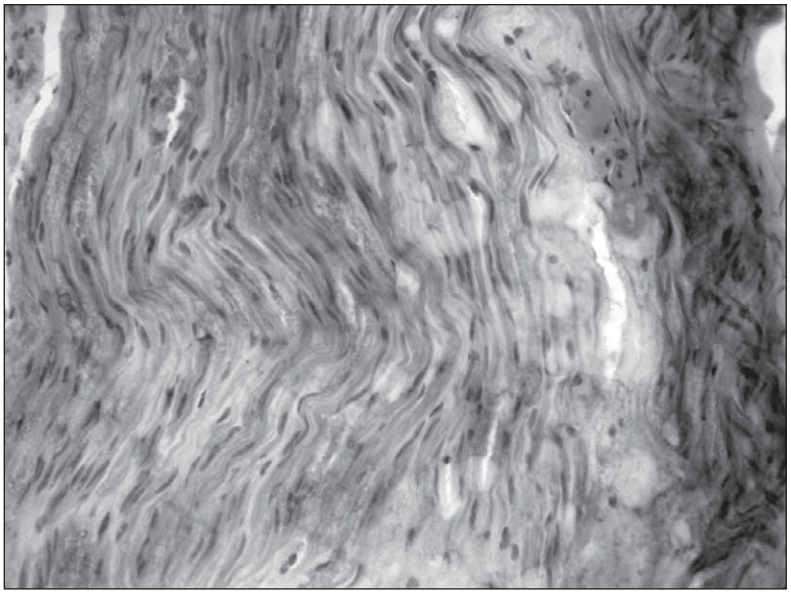

FIGURE 2. *Perivascular unstructured material in the endoneurium. HE,ob.40.*

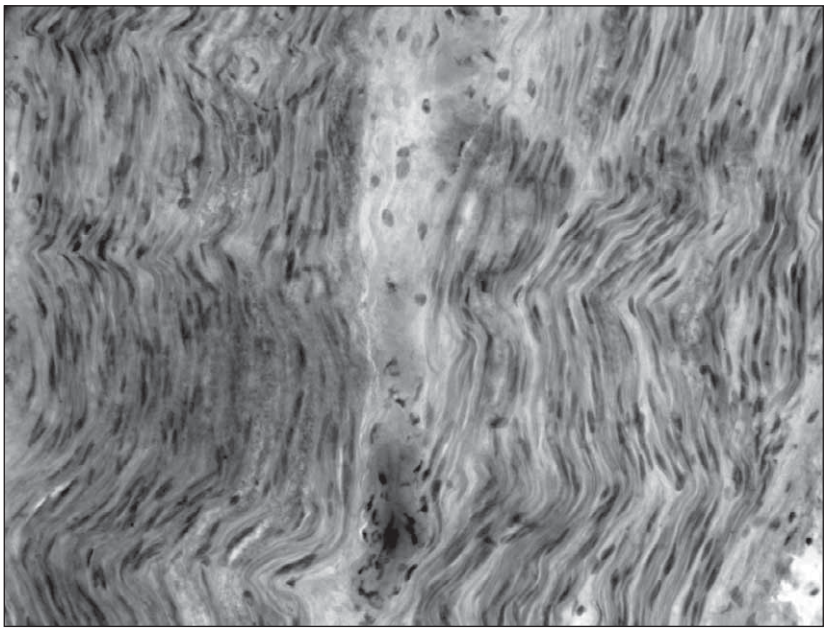

FIGURE 3. *Amyloid deposits along the vessels in the endoneurium. Congo red, ob.40.*

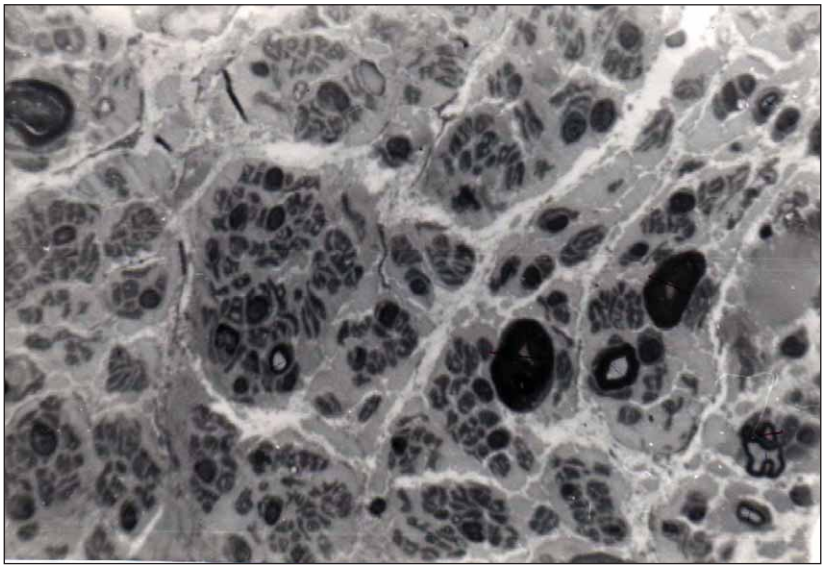

FIGURE 4. *Severe myelinic fiber loss, myelinic bullae, amyloid deposits. SSF, ob.100*

The electron microscopy (figs.7 and 8) confirmed the amyloid origin of the unstructured stored material, the important collagen proliferation, the presence of Bungner bands, collagen pockets and the extremely small number of amyelinic axons explaining the impossibility to perform their histogram.

Teased myelinic fiber examination showed: 72% normal fibers, 8% fibers with axonal degeneration lesions, 10% fibers with segmental demyelination lesions and 10% fibers with remylination signs.

The conclusion of the sural nerve biopsy was amyloid neuropathy: chronical severe interstitial neuropathy involving especially amyelinic fibers; primary lesions: axonal-demyelinating lesions with important endoneurial amyloid deposits.

Gastrocnemius muscle biopsy showed important neurogenic lesions.

After more than 10 years we performed sural nerve and gastrocnemius muscle biopsies for the daughter of the patient we present now. She was 20 years old and had the clinical signs of a polyneuropathy predominantly autonomic. Her nerve biopsy showed lesions of mixed axonal-demyelinating neuropathy without amyloid deposits, at least in the nerve fragment sampled, probably because of the focal type of the lesions which characterize the

FIGURE 5. *Amyloid deposits around the vessels. SSF, ob.100*

amyloid neuropathy, or may be to the young age of this patient.

Unfortunately molecular genetic tests could not be performed for our patient, that is why the differential diagnosis among the different types of amyloidoses associating peripheral neuropathy had to be based only on clinical and general paraclinical data of the patient.

Our presentation is in fact an opportunity to review the nowadays knowledge in this field.

It is generally accepted that the diagnosis of an amyloidosis needs the biopsy of an affected organ: nerve, kidney, etc. or a biopsy of rectal mucosa, gum, salivary gland, etc. In most cases the peripheral nerve biopsy is the most useful although, sometimes, like in the daughter of our patient, the amyloid deposits can be absent in the nerve fragment examined. In such situation the association of different clinical signs must be considered.

The most frequent form of hereditary systemic amyloidosis with autosomal dominant inheritance is the transthyretin (TTR) amyloidosis (Andrade, 1952).

Transthyretin (TTR) is a plasmatic protein transporting the tyroidian hormone and a retinol-binding protein/vitamin A (Robbins, 1976). TTR is synthetised in the liver and is expressed also in the choroids plexus and in the pigmentary retinian epithelium.

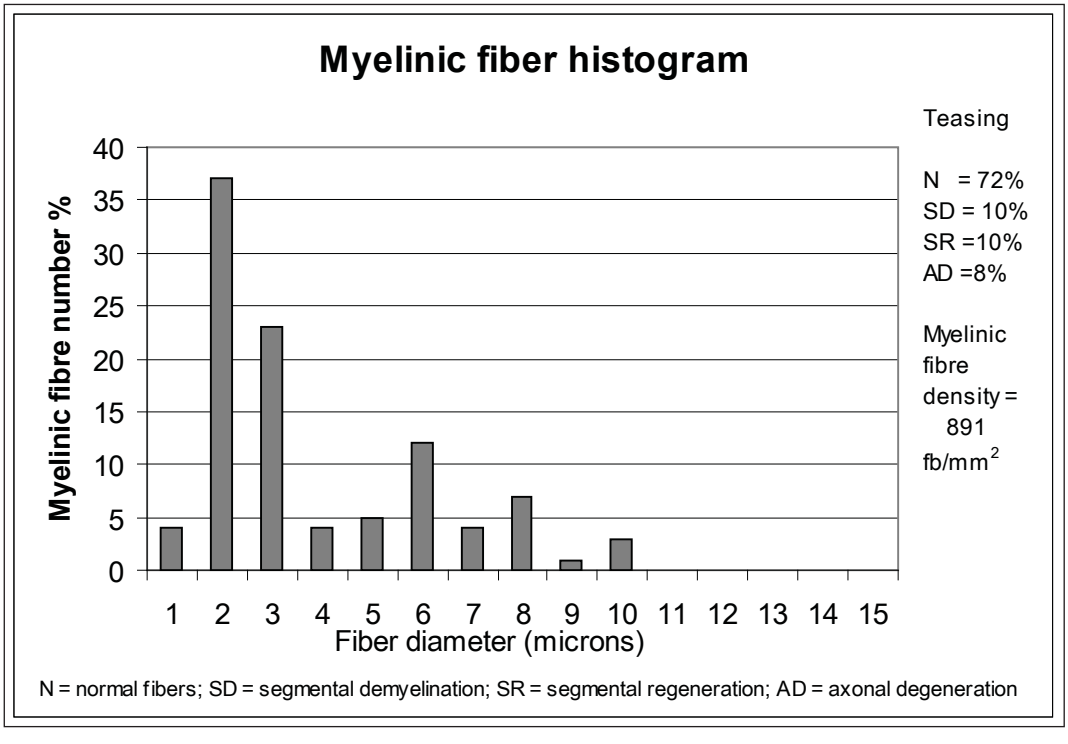

FIGURE 6. *Myelinic fiber histogram and teasing fiber data.*

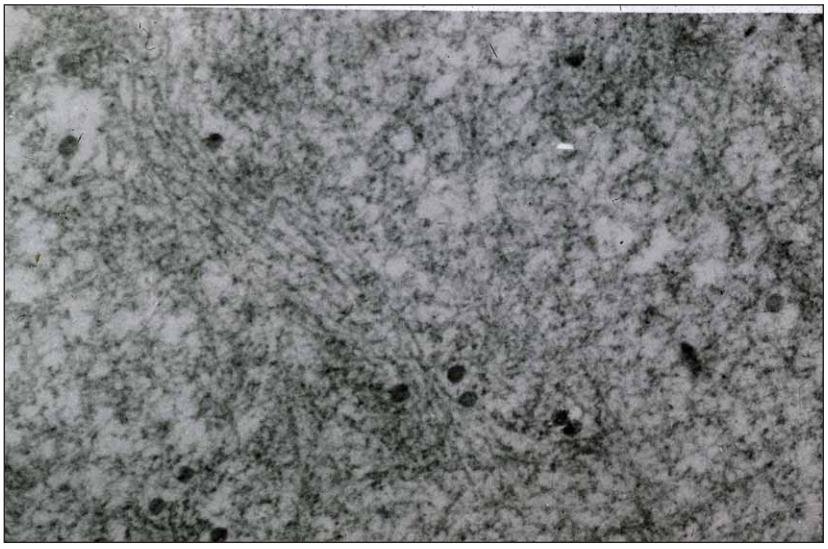

FIGURE 7. *Amyloid deposits. EM.*

FIGURE 8. *Collagen proliferation, Bungner bands, collagen pockets.EM.*

Hundreds of mutations in the TTR gene (chromosome 18) are known, most of them being associated with amyloidosis (Benson et al, 1996; Connors et al, 2003). The penetrance and the clinical phenotype of the disease depend upon the mutation, but also upon the family involved: Swedish and Portuguese families have the same mutation but different clinical phenotypes.

The clinical features of TTR amyloidosis is very variable, but the sensorimotor polyneuropathy is the most frequent, reason for which the disease was long time named familial amyloid polyneuropathy (FAP). The onset of the disease is usually insidious with distal bilateral thermo+/- algic dysesthesia with ascendant progression in the lower limbs,then superior limbs too, motor dysfunction appearing later.

Autonomic dysfunction appears early its signs being sometimes the first: sexual impotence, ortostatic arterial hypotention, bladder and gastrointestinal dysfunctions.

Electrophisiological studies (nerve conduction velocities) are very useful (Sales, 1978; Said et al, 1984). Among other benefits electrophysiological studies help the differential diagnosis between the polyneuropathy from the carpal tunnel, both appearing in different proportions more or less in relation with the mutation type.

Among other signs of the TTR amyloidosis we mention: the laryngeal nerve involvement, the pathognomonic pupil deformation named "scalopped pupil", vitrous opacities, cardiomyopathy and the oculoleptomeningeal syndrome: stroke or seazures or hydrocephalus, or medulary infarct, or cerebral hemorrhage in late stagies (Benson and Kinkaid, 2007).

Nowadays death is due to the cardiomyopathy. In the past, death was due to leg infected trophic lesions, osteomyelitis or malnutrition.

Concerning the pathogeny of TTR amyloidosis, it is known that transthyretin, the normal one included, having a beta-pleated structure tends to form fibrils.TTR catabolism depends on the mutation type and may contribute to the fibril formation and deposition. Details concerning this subject can be found in the paper of Benson and Kinkaid (2007).

Although the amyloid deposits in the vessel walls are spreadeverywhere in the body, in TTR amyloidosis the liver and the spleen are preserved. In the peripheral nerve the amyloid deposition begin around the arterioles at random, that is why small amyloid deposits may escape to the examination of a biopsy. Progressively, globular deposits appear in the endoneurium as well as the demyelinating-axonal neuropathy morphological signs like in our case.

Although TTR amyloidosis may not be excluded in our patient, this probability is reduced because he had no clinical signs from other organs, he had no nephritic syndrome and no familial history, but a "de novo" mutation would be possible.

Another type of hereditary amyloidosis with autosomal dominant transmission possibly associated with peripheral neuropathy is AI alipoprotein amyloidosis (ApoAI) described by Van Allen et al (1969). Twelve mutations in the ApoAI gene (chromosome 11) associated with the systemic amyloidosis are known, but only Gly26Arg mutation may be associated with a polyneuropathy, and not always. Details concerning the pathogeny and genetics of this disease are reported by Nichols et al (1990), Rader et al (1992) and Borhani et al (1997).

In the ApoAI amyloidosis the first and most important clinical manifestation is the azothemia due to amyloidotic lesions of the renal medulary vessels contrary to the proteinuria due to renal glomerulopathy lesions in the primary and secondary amyloidoses. Proteinuria is absent in ApoAI as well as in TTR amyloidoses. In the last one the amyloid deposits in the kidney are not quantitatively sufficient to produce a renal insufficiency.

ApoAI polyneuropathy is clinically and morphologically similar to the polyneuropathy in the TTR amyloidosis, but motor dysfunction is much more severe: weakness in the leg muscles has an early onset as well as foot drop and amyotrophy. Ataxia and the deep tendon reflexe loss are common signs. Patients may become tetraparetic. Impotence is a common feature in man. Large amyloid deposits have been reported in dorsal root ganglia. Some amyloid deposits may appear in spinal and cerebral leptomeninges too, but like in TTR amyloidosis cerebral parenchyma is spread and vitrous opacities are absent.

In our patient the absence of ataxia and the reduced motor dysfunction exclude ApoAI amyloidosis. Of course the clinical phenotype of a disease is not always completely expressed in a patient, but the absence of azotemia is a major argument in this case.

The clinical features in our patient are different from those known in the third type of hereditary amyloidosis associated with peripheral nerve involvement: gelsolin amyloidosis (AGel) due to mutations in gelsolin gene – an actin-modulatory protein. Mutations in Asp187Asn have been reported by Levy et al (1990) and Maury et al (1990) and in Asp187Tyr by de la Chapelle et al (1992).

Gelsolin amyloidosis is a rare disease with adult onset, clinically characterized by a corneal dystrophy followed by a progressive cranial nerve neuropathy and later by a peripheral neuropathy (Meretoja and Teppo, 1971). Other organ involvement is reduced. The diagnosis of gelsolin amyloidosis is clinically excluded in our case by the absence of cornea and cranial nerve involvement.

In primary amyloidosis or light chain immunoglobulin amyloidosis (AL), a sporadic form of amyloidosis, 20% of the patients have a peripheral polyneuropathy. The nerve biopsy can not help the differential diagnosis from the hereditary forms, the morphological aspects of the nerve being similar in all of them.

The diagnosis of AL amyloidosis is sustained by the presence of a plasmocytic discrasia or by the presence of a monoclonal immunoglobulinemia or immunoglobulinuria (80% of cases) (Kyle and Gertz,1990).

The benign monoclonal IgG gammopathy in our case could suggest a sporadic primary amyloidosis, but his daughter had the same type demyelinating-axonal neuropathy with nonproved amyloid.

The utility of genetic tests in such cases is evident, although the discovery of a new unknown mutation may offer surprises and necessite extended studies. Genetic studies are expensive and specialized laboratories are necessary. DNA tests for ApoAI and AGel amyloidoses, rare diseases, are not yet available.

A secondary amyloidosis is to be excluded from the differential diagnosis of our patient disease, the

personal history of our patient being incompatible with a secondary amyloidosis. Moreover, cases of peripheral neuropathy associated with a secondary amyloidosis have not been reported.

Family history may be useful but insufficient for the differential diagnosis due to reduced disease penetrance, variability of the clinical phenotype and the possibility of a "de novo" mutation (Benson and Kinkaid, 2007).

The amyloid neuropathy therapy comprises symptomatic therapies against neuropathic pain, digestive, urinary and erectile dysfunctions. Holmgren et al (1993) recommend liver transplant in TTR amyloidosis, although it was proved that amyloid deposition continues. Only Ikeda et al (2003) reported amelioration of neuropathic signs after liver transplant. Hammarstrom et al (2003) and Razavi et al (2003) recommend diflumisol – a nonsteroidal antiinflammatory drug in TTR amyloidosis whose small molecules bind and stabilize thermodynamically TTR.

Very recently suppression of liver mutant TTR synthesis by antisens oligonucleotides (Benson et al (2006) was experimentally studied in mice. Another therapy was proposed by Tanaka et al in 2001 utilizing rybozimes that suppress TTR expression in hepatic cell cultures.

Conclusions on amyloidosis. Vitrectomy has a transitory effect and may be followed by glaucoma and retinal detachment. Oculoleptomeningeal syndrome has no treatment till now. For AGel amyloidosis only corneal transplant, possibly repeted, may give amelioration. Studies concerning different aspects of systemic amyloidoses continue.

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